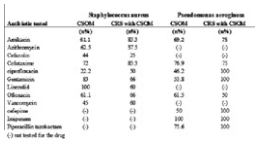

Conclusion: The antibiotic sensitivity of the common microbes differed significantly between CSOM and CRS with CSOM subjects in South Indian Population. The present study warrants the need for evaluation of antimicrobial susceptibility profile of the causative microbial pathogens before administration of antibiotics to treat CRS with CSOM in particular.

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The emergence of cotrimoxazole and quinolone resistance in Shigella sonnei

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Background: The emergence of cotrimoxazole resistance has been a dominant and consistent character in our isolates of Shigella sonnei. To study the behaviour of these emerging strains and characterise mechanisms of resistance to cotrimoxazole & ciprofloxacin the following study was performed.

Methods & Materials: Isolates of *Shigella sonnei* confirmed by standard methods from 2012 to 2015 were subjected to antimicrobial susceptibility testing using the Kirby Bauer method as per Clinical laboratory standards institute and PCR for the detection of virulence genes. The degree of relatedness between the isolates was assessed by ERIC PCR followed by gel image analysis. Dendrogram was generated using Pyelph. PCR was carried out to determine the mechanisms of resistance to cotrimoxazole and ciprofloxacin.

Results: Of 34 *Shigella sonnei* isolates, cotrimoxazole resistance was common (94.1%) followed by ciprofloxacin (47%). Majority carried the *ipaH* gene (97%) followed by *ial* (17.6%), *sen* (11.7%), *set 1* & *set2* (5.8%). No *stx* element was found. ERIC–PCR analysis of the isolates resulted in four major ERIC groups labelled Eric group I,II,III and IV. Type III was the dominant (44.1%) type. Majority harboured *dhfr1* (94.1%), *sul2* (85.2%) followed by *sul3* (55.8%), *sul1* (11.7%). Two isolates that were resistant to cotrimoxazole were negative for the *sul* genes but harboured the *dhfr1* gene. All the phenotypically ciprofloxacin resistant isolates (47%) were positive for presence of *gyr A,gyr B, parC and parE*. Also, *qnrB* was the most prevalent PMQR gene (93.7%) while, *qnrC* was positive in 18.7% of isolates. None were positive for *qnrA and qnrS*. Two (0.1%) of the isolates were positive for *aac(6')-lb* gene. The *qepA* gene regulating the efflux pump was negative in all the isolates studied. One isolate that was susceptible to all antibiotics tested negative for all the genes.

Conclusion: The study revealed presence of gene cassette mediated *bla*$_{ESBLs}$ among gram negative rods within hospital environment. Presence of *bla*$_{ESBLs}$ in diverse Inc plasmid groups suggests their diverse source of acquisition. Current study insists vital detail and the increasing trend of resistance to quinolones is a point of concern. This study also shows the emergence of a particular ERIC type in the background of this evolving resistance pattern.

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Carriage of multiple gene cassettes mediated extended spectrum cephalosporinase within diverse incompatibility (Inc) plasmid groups among gram negative rods in a tertiary referral hospital of India

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Background: Extended spectrum β-lactamases pose to be major health problem in hospital settings worldwide. Infection with ESBL producing organisms result in poor clinical outcome, overdue initiation of suitable antibiotic treatment, longer hospital stays and greater hospital operating cost. Management of treatment against these strains become complicated when the resistant determinants are associated with integron and horizontally transferable due to their location within plasmid. In this study, we report multiple gene cassettes mediated extended spectrum cephalosporinase within diverse Inc plasmid groups among gram negative rods for the first time in India.

Methods & Materials: A total number of 458 clinical isolates of gram negative rods were collected during November 2011 to October 2013 from Silchar Medical College and Hospital. ESBL status was detected by phenotypic screening as per CLSI criteria and multiplex PCR assay followed by sequencing. Genetic environment was determined by integrase gene PCR and location of *bla*$_{ESBLs}$ within gene cassette was investigated by 59base element PCR and sequencing. Plasmid transferability was done by transformation and conjugation while incompatibility profiling was done by PCR based replicon typing. DNA finger printing of isolates was done by ERIC and REP PCR.

Results: A total of 56 isolates were found harboring *bla*$_{ESBLs}$ by PCR and sequencing. All of them were carrying class I integron and *bla*$_{ESBLs}$ was found to be located within gene cassette and conjugative plasmid. Further, PCR based replicon typing established presence of diverse Inc plasmid types viz. FIA, FIB, P, F$_{rep}$B, K, B/O, I1/Iγ and Y. The isolates showed high MICs against cephalosporins (≥256 μg/ml), and monobactam (≥256 μg/ml) but was found in susceptible range against Ertapenem drug. The isolates were found clonally unrelated.

Conclusion: The study revealed presence of gene cassette mediated *bla*$_{ESBLs}$ among gram negative rods within hospital environment. Presence of *bla*$_{ESBLs}$ in diverse Inc plasmid groups suggests their diverse source of acquisition. Current study insists vital

requirement for regular monitoring of these resistant determinants to execute the right antibiotics policy so as to reduce the irrational use of expanded spectrum cephalosporins and to decrease the antibiotic pressure and treatment failure in clinical setting in this part of the world.

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Beta-lactamases in a Nepalese hospital: Wake up before the "biological quake" destroys you

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Background: In this era of modern medicine, antimicrobial resistance has emerged as a major health catastrophe. Emergence of drug resistance mechanisms like Extended-spectrum beta–lactamases (ESBLs), AmpC beta-lactamases and metallo-beta-lactamases (MBLs) can be regarded as "biological quake" posing therapeutic challenge to the health care settings. Therefore, this study was designed to determine the prevalence of ESBL-, MBL-, AmpC-producing bacteria in hospital-admitted patients.

Methods & Materials: A prospective study was conducted among the inpatients of Medicare National Hospital in central Kathmandu for four months (April-July, 2015), a period when the hospital was engaged with "Nepal Earthquake 2015" victims too. Different clinical specimens were collected, processed and the isolates were identified following standard methodology. Antibiotic sensitivity test was done by Kirby-Bauer disc diffusion method. ESBL was detected by standard combination disc method. Besides, tests for ESBL, AmpC, and co-production of ESBL and AmpC were done by MASTDISCS™ ID AmpC and ESBL Detection Discs, as well as ESBL and AmpC detection Ezy MIC™ Strip (HiMedia, India). EDTA-Imipenem combination disc method was followed for MBL detection.

Results: Among the total 75 gram-negative bacterial isolates resistant to third generation cephalosporin, ESBL was seen in 30.6% (n=23). Similarly, MBL and AmpC production were seen in 8% (n=6) and 1.3% (n=1) respectively. Interestingly, ESBL-AmpC co-production was found in 4% (n=3). *Escherichia coli* was the most frequent ESBL-producer (n=20). *E. coli* was found to produce MBL (n=4), AmpC (n=1), and ESBL-AmpC combination (n=2) as well. Two isolates of *P. aeruginosa* were ESBL-AmpC co-producers. Out of 23 ESBL-producer, 78.2% (n=18) were from intensive care unit patients. The ESBL-producing bacteria showed sensitivity to different antibiotics as follows- meropenem (n=21, 91.3%), amikacin (n=20, 86.9%), and cefoperazone-sulbactam (n=19, 82.6%). Consistent results were found with different methods employed for detection of ESBL and AmpC.

Conclusion: ESBL-producing bacteria were more commonly seen though AmpC- and MBL-producers were relatively less frequent. Special strategy like antibiotic stewardship should be followed in our setting before the situation turns out to be havoc. Identification, characterization and surveillance of antibiotic susceptibility profile of beta-lactamase-producing organisms can lead to successful infection control.

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Fecal carriage of carbapenem resistant enterobacteriaceae (CRE) and risk factor analysis in hospitalised patients: A single centre study from India

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Background: Carbapenem resistant *Enterobacteriaceae* (CRE) have emerged and disseminated widely causing a variety of infections. The emergence of carbapenem resistant *Enterobacteriaceae* is associated with limited therapeutic options and increased mortality in patients infected by these strains. These organisms also have the propensity to undergo widespread dissemination via mobile genetic elements. Enteric strains possessing these carbapenemases have shown remarkable success in the form of large scale geographical dissemination. Gut colonization by CRE may act as reservoir of these pathogens for dissemination within an enclosed setting as in a hospital. To the best of our knowledge, there are no studies of CRE fecal carriage using genotypic methods and those analysing risk factors leading to such colonization in hospitalised patients in India.

Methods & Materials: We conducted the present study to observe gut carriage rate of CRE in patients presenting to our tertiary care hospital using both phenotypic (modified Hodge test) and genotypic (polymerase chain reaction for bla_{VIM}, bla_{KPC},bla_{IMP} and bla_{NDM-1} genes) methods and tried to identify the risk factors for CRE gut colonization.

Results: A total of 239 fecal swabs yielded 259 *Enterobacteriaceae* isolates, of which 108 isolates (majority included *E. coli* and *Klebsiella* spp.) from 84 patients showed presence of CRE (prevalence 84/239; 35.14%); 28 isolates from 23 patients had bla_{NDM-1} while 20 isolates from 17 patients possessed bla_{VIM} gene. No isolate was positive for bla_{KPC} and bla_{IMP} genes. Although highest isolation of CRE was from the wards, approximately half of patients of intensive care units yielded CRE in fecal swabs. The CRE were also found to have significantly high antimicrobial resistance as compared to non-CRE isolates. Multivariate analysis of risk factors showed use of any antibiotic (P = 0.002), cephalosporins use (P = 0.000) and presence of any indwelling device (P = 0.014) as independent risk factors for acquiring gut colonization.

Conclusion: The study is the first from India to show high CRE carriage in patients admitted to a tertiary care centre and emphasises the need of strict antimicrobial stewardship implementation in hospitals to prevent dissemination of multidrug resistant CRE.

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